Paroxysmal Nocturnal Haemoglobinuria

Participant: 42981 **Trial No: 151601**

Date Issued: 18-May-2015 **Closing Date: 06-June-2015**

Your Results

Sample: 077

Trial Comments

This trial was issued to 156 participants.

Sample Comment

The sample was manufactured by UK NEQAS using a sample of blood from a PNH patient and a stabilised whole blood unit

Target Cells	Your Result	Median Result	Lower Quartile	Upper Quartile	Number of Returns
RBC PNH Clone - CD59		3.65	2.63	5.02	124
RBC Type II Cells - CD59		0.40	0.26	0.60	113
RBC Type III Cells - CD59		3.06	2.18	4.36	114
Granulocytes PNH Clone - CD16		15.50	13.95	16.62	35
Granulocytes PNH Clone - CD24		15.00	14.30	16.00	101
Granulocytes PNH Clone - CD55		15.05	13.78	16.25	20
Granulocytes PNH Clone - CD59		14.20	12.40	15.85	19
Granulocytes PNH Clone - CD66b		15.70	14.19	16.47	31
Granulocytes PNH Clone - FLAER		15.10	14.18	16.21	124
Granulocytes PNH Clone - CD157		14.89	14.02	16.18	10

Performance Monitoring

Sample 077

	Consensus	Your Result	Classification	Running Classification
RBC	Clone Present		Outwith Consensus	Red [3]
WBC	Clone Present		Outwith Consensus	Red [3]

1 = All Samples Within Consensus across last 3 samples
2 = 1 Sample Outwith Consensus across last 3 samples
3 = 2 or more Samples Outwith Consensus across last 3 samples
= Not enough samples to determine Running Consensus

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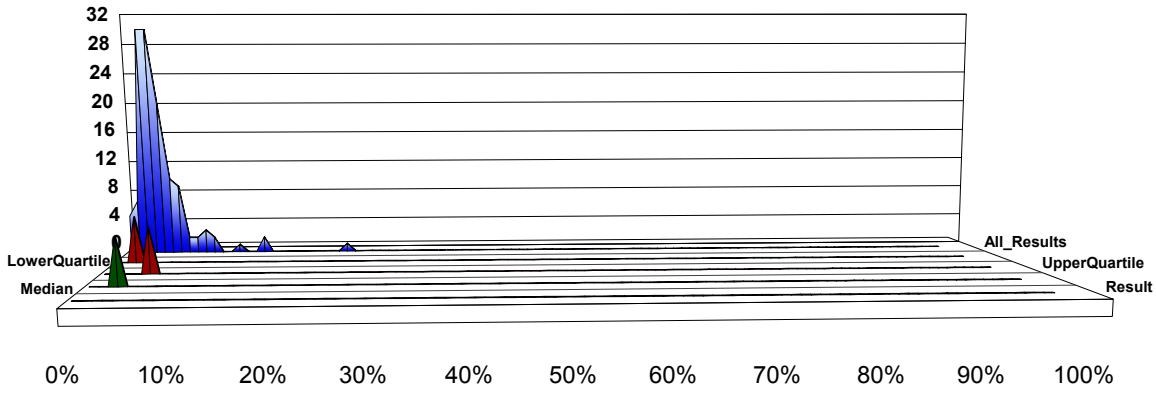
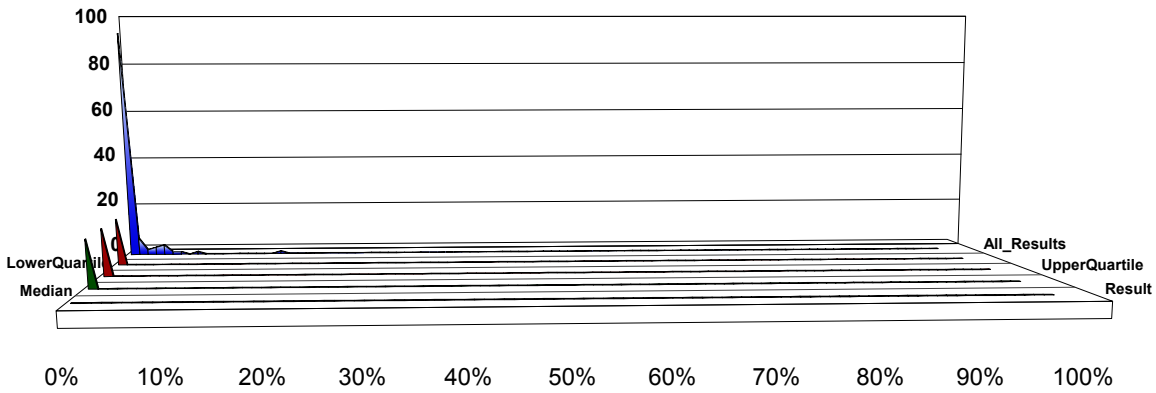
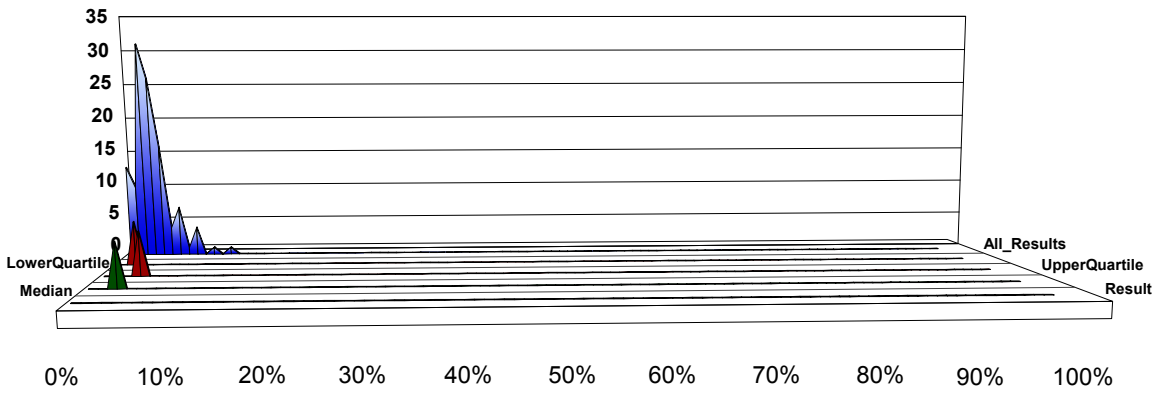
RBC PNH Clone - CD59



RBC Type II Cells - CD59



RBC Type III Cells - CD59



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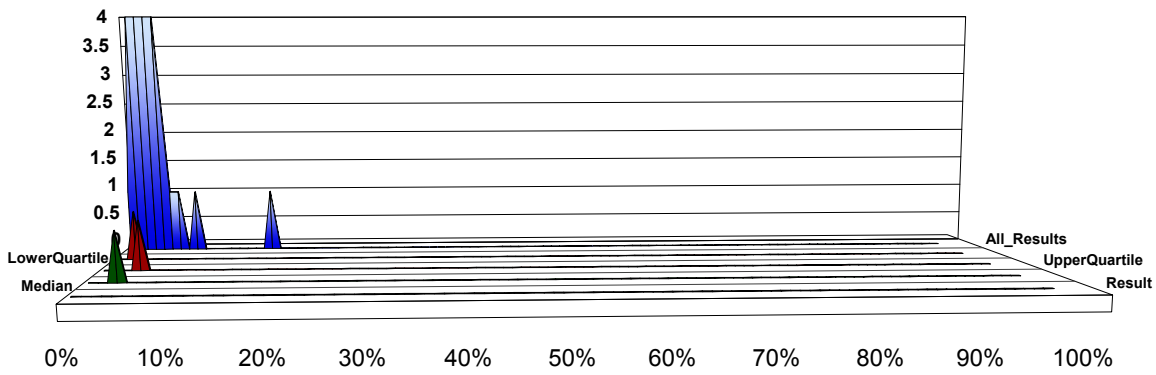
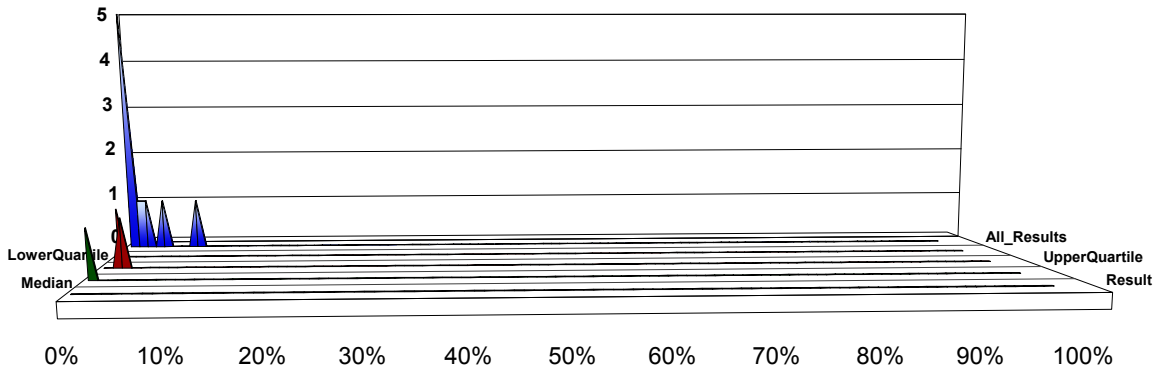
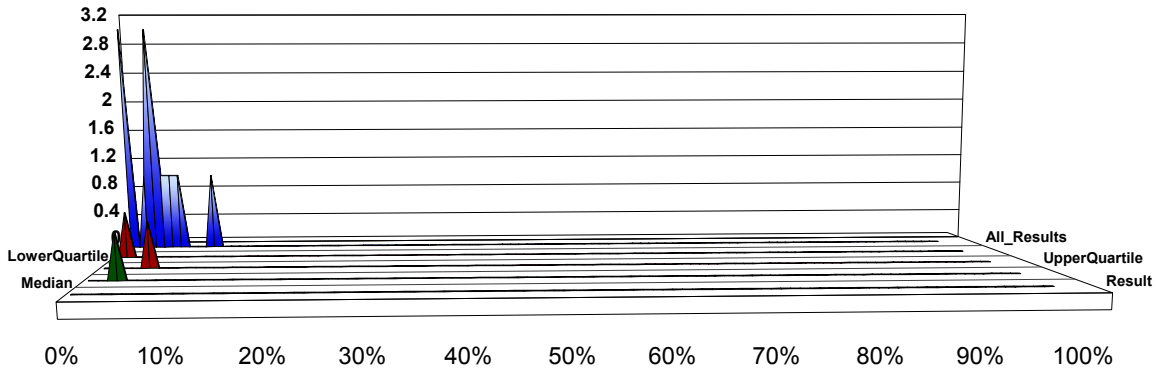
RBC PNH Clone - CD55



RBC Type II cells - CD55



RBC Type III cells - CD55



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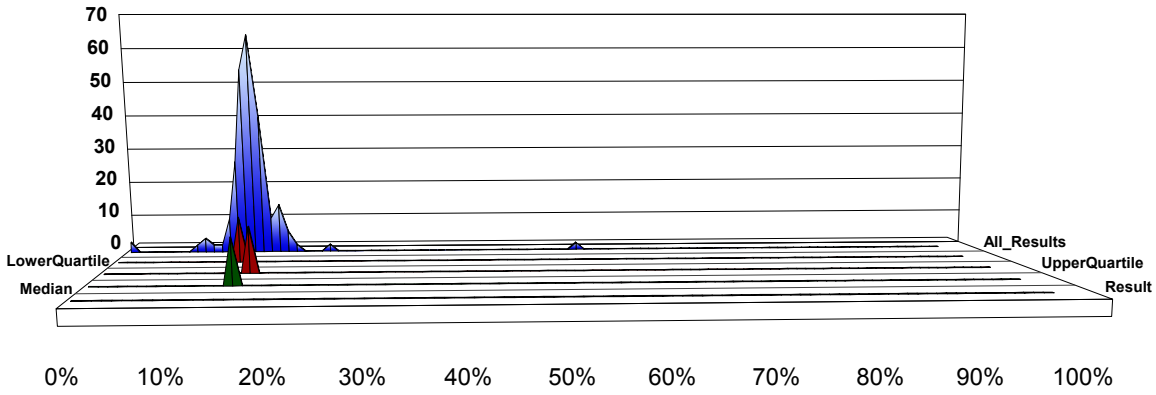
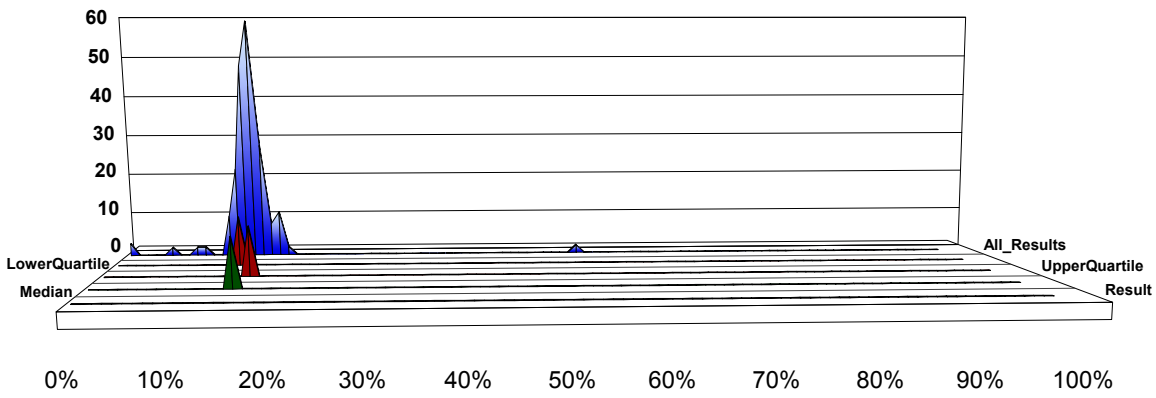
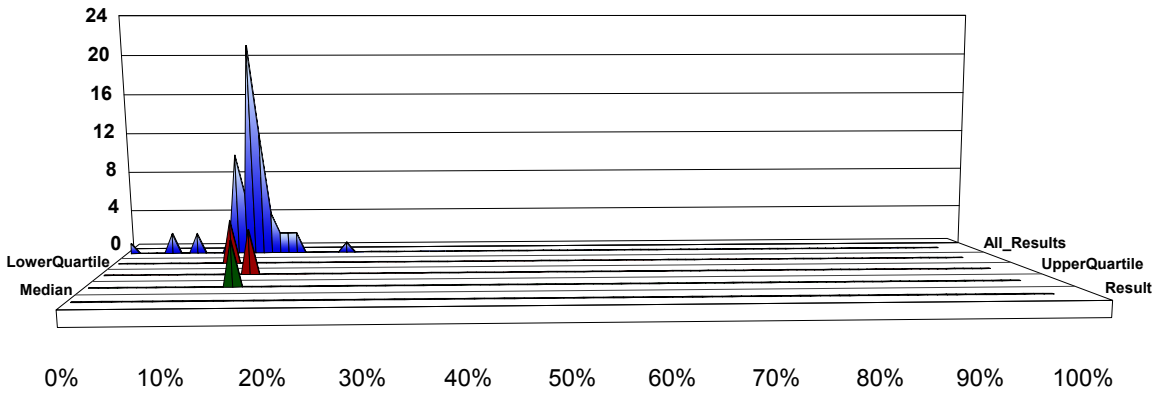
Granulocytes PNH Clone - FLAER



Granulocytes PNH Clone - CD24



Granulocytes PNH Clone - CD16



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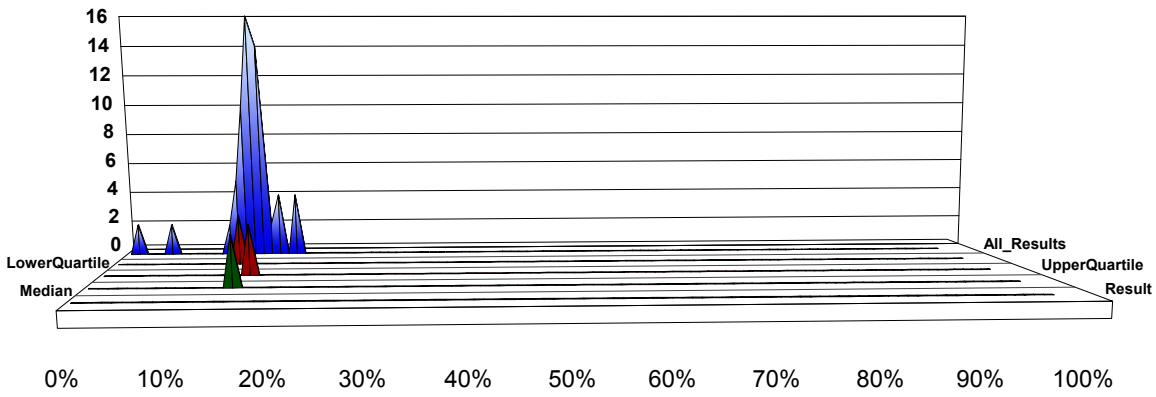
Granulocytes PNH Clone - CD66b



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Method Breakdown

RBC PNH Clone

Flow Cytometer	Median	Lower Quartile	Upper Quartile	Returns
CD55				
All	3.30	2.50	4.70	21
BD	3.20	1.92	4.90	17
CD59				
All	3.65	2.63	5.02	124
BD	3.80	2.67	5.11	87
Coulter	3.42	2.60	4.55	36

Antibody Manufacturer	Median	Lower Quartile	Upper Quartile	Returns
CD55				
All	3.30	2.50	4.70	21
BD Biosciences	2.90	1.88	4.15	11
CD59				
All	3.65	2.63	5.02	124
BD Biosciences	3.25	2.43	4.64	14
Beckman Coulter	3.53	2.15	4.15	11
eBioscience	4.72	3.49	6.63	10
invitrogen	3.60	2.63	4.54	60

Fluorochrome	Median	Lower Quartile	Upper Quartile	Returns
CD55				
All	3.30	2.50	4.70	21
PE	3.05	1.90	4.89	16
CD59				
All	3.65	2.63	5.02	124
PE	3.74	2.68	5.05	94
FITC	3.53	2.57	4.80	29

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Method Breakdown
Granulocytes PNH Clone

Flow Cytometer	Median	Lower Quartile	Upper Quartile	Returns
CD157				
All	14.89	14.02	16.18	10
CD16				
All	15.50	13.95	16.62	35
BD	15.30	14.10	16.50	25
CD24				
All	15.00	14.30	16.00	101
BD	15.00	14.33	15.75	67
Coulter	15.00	13.92	16.49	33
CD55				
All	15.05	13.78	16.25	20
BD	15.05	13.78	16.88	12
CD59				
All	14.20	12.40	15.85	19
BD	13.23	10.70	14.37	12
CD66b				
All	15.70	14.19	16.47	31
BD	15.40	14.39	16.22	22
FLAER				
All	15.10	14.18	16.21	124
BD	15.08	14.20	16.05	83
Coulter	15.33	13.89	16.31	40

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Antibody Manufacturer	Median	Lower Quartile	Upper Quartile	Returns
CD157				
All	14.89	14.02	16.18	10
CD16				
All	15.50	13.95	16.62	35
BD Biosciences	15.56	15.00	16.64	13
Beckman Coulter	15.90	14.55	17.00	13
CD24				
All	15.00	14.30	16.00	101
BD Biosciences	15.04	14.40	15.92	26
Beckman Coulter	14.90	13.96	16.26	47
Pharmingen	15.05	14.55	15.83	20
CD55				
All	15.05	13.78	16.25	20
BD Biosciences	14.73	13.43	15.95	10
CD59				
All	14.20	12.40	15.85	19
CD66b				
All	15.70	14.19	16.47	31
BD Biosciences	16.60	15.00	18.40	13
FLAER				
All	15.10	14.18	16.21	124
Other	15.26	14.38	16.23	69
Protox	15.00	14.20	16.18	46

Fluorochrome	Median	Lower Quartile	Upper Quartile	Returns
CD157				
All	14.89	14.02	16.18	10
CD16				
All	15.50	13.95	16.62	35
CD24				
All	15.00	14.30	16.00	101
PE	15.10	14.10	16.14	73
CD55				
All	15.05	13.78	16.25	20
PE	15.00	14.00	17.00	13
CD59				
All	14.20	12.40	15.85	19
PE	14.30	13.27	16.73	12
CD66b				
All	15.70	14.19	16.47	31
FITC	15.30	14.20	16.65	23
FLAER				
All	15.10	14.18	16.21	124
FITC	15.10	14.00	16.00	33
AlexaFluor 488	15.24	14.23	16.36	90

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Paroxysmal Nocturnal Haemoglobinuria

Participant: 42981 **Trial No: 151601**

Date Issued: 18-May-2015 **Closing Date: 06-June-2015**

Your Results

Sample: 078

Trial Comments

This trial was issued to 156 participants.

Sample Comment
The sample was manufactured by UK NEQAS using a stabilised whole blood unit

Target Cells	Your Result	Median Result	Lower Quartile	Upper Quartile	Number of Returns
RBC PNH Clone - CD59		0.00	0.00	0.00	122
RBC Type II Cells - CD59		0.00	0.00	0.00	111
RBC Type III Cells - CD59		0.00	0.00	0.00	113
Granulocytes PNH Clone - CD16		0.00	0.00	0.00	35
Granulocytes PNH Clone - CD24		0.00	0.00	0.00	97
Granulocytes PNH Clone - CD55		0.00	0.00	0.07	19
Granulocytes PNH Clone - CD59		0.00	0.00	0.00	20
Granulocytes PNH Clone - CD66b		0.00	0.00	0.00	30
Granulocytes PNH Clone - FLAER		0.00	0.00	0.00	122
Granulocytes PNH Clone - CD157		0.00	0.00	0.00	10

Performance Monitoring

Sample 078

	Consensus	Your Result	Classification	Running Classification
RBC	Clone Absent		Outwith Consensus	Red 3
WBC	Clone Absent		Outwith Consensus	Red 3

1 = All Samples Within Consensus across last 3 samples
2 = 1 Sample Outwith Consensus across last 3 samples
3 = 2 or more Samples Outwith Consensus across last 3 samples
= Not enough samples to determine Running Consensus

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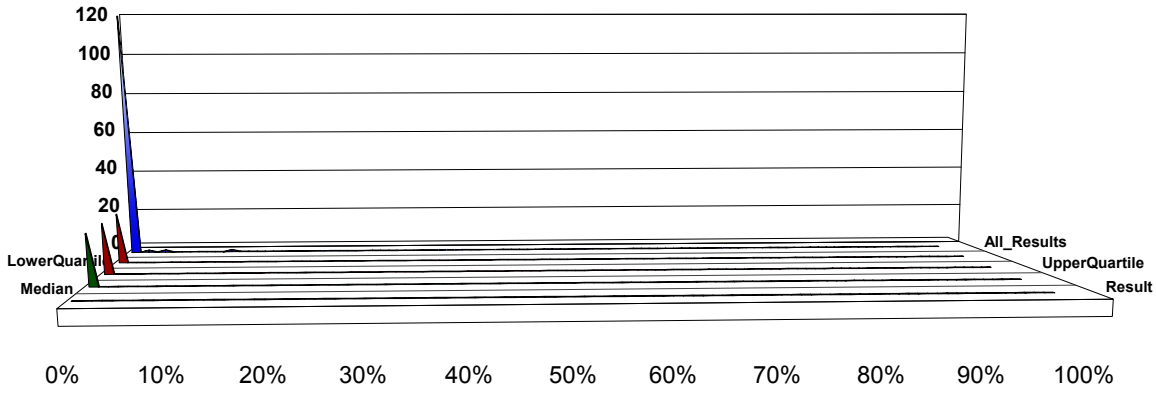
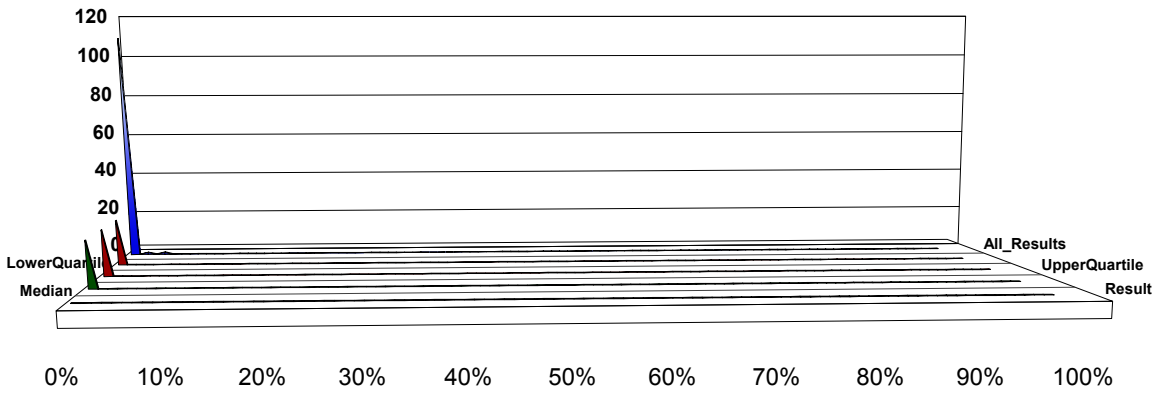
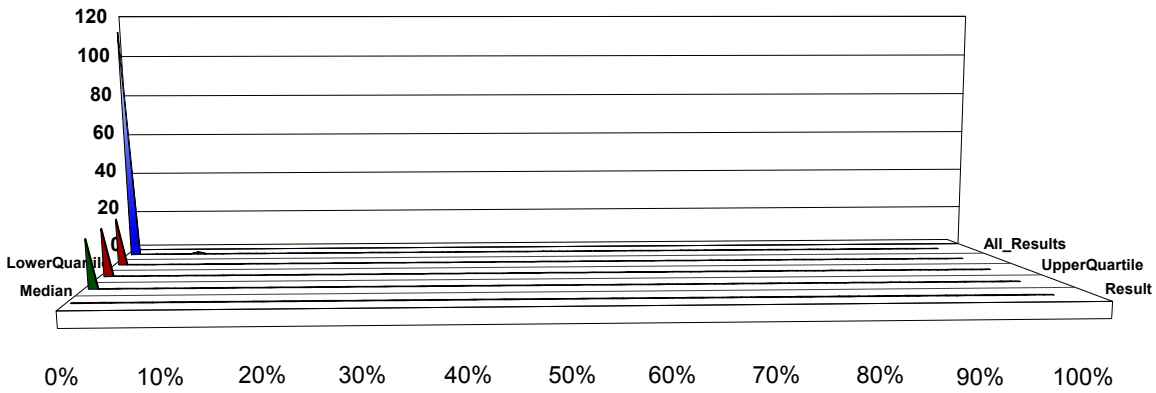
RBC PNH Clone - CD59



RBC Type II Cells - CD59



RBC Type III Cells - CD59



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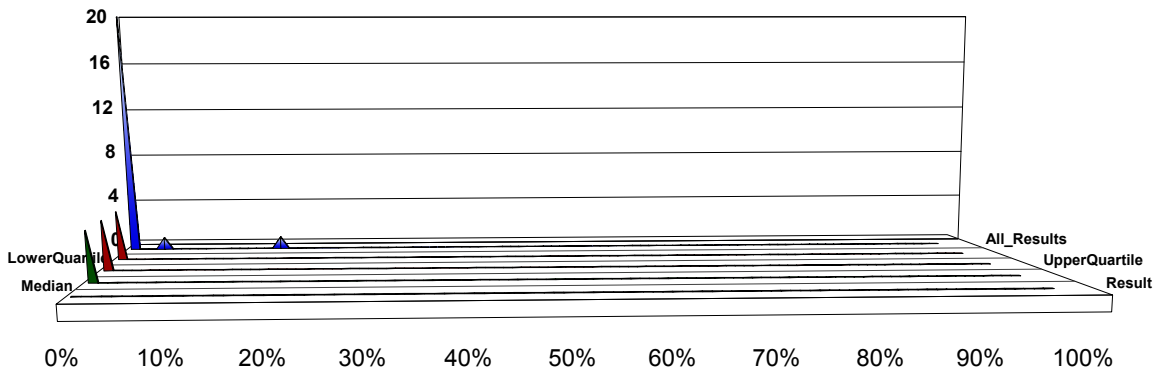
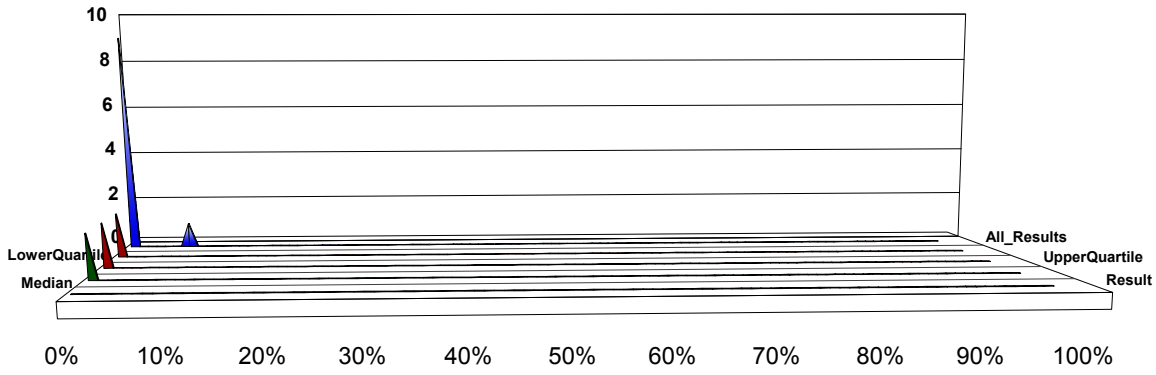
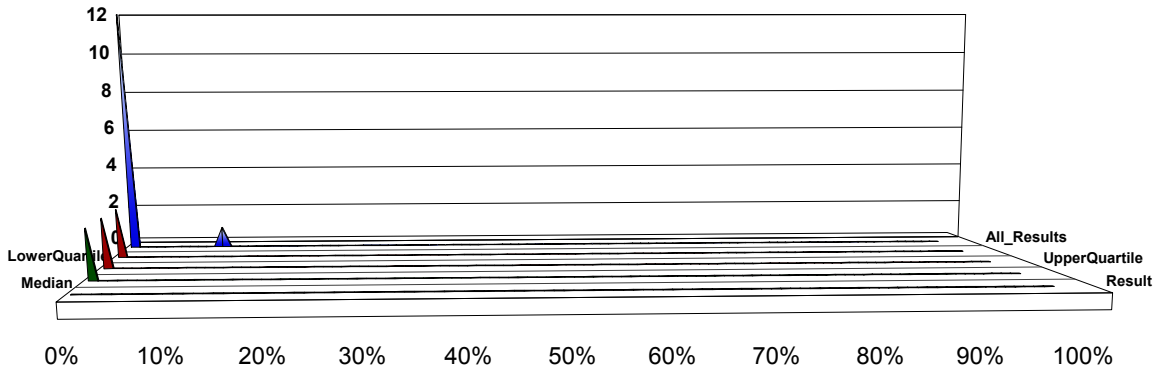
RBC PNH Clone - CD55



RBC Type II cells - CD55



RBC Type III cells - CD55



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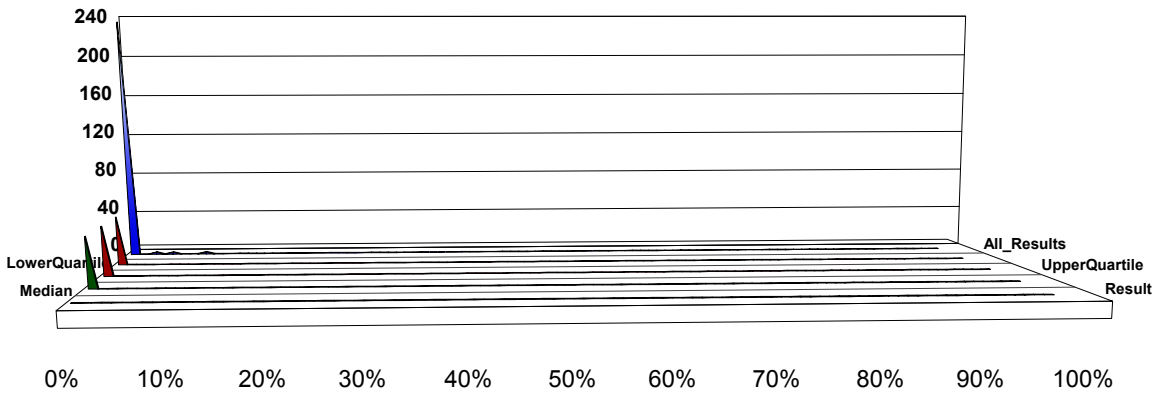
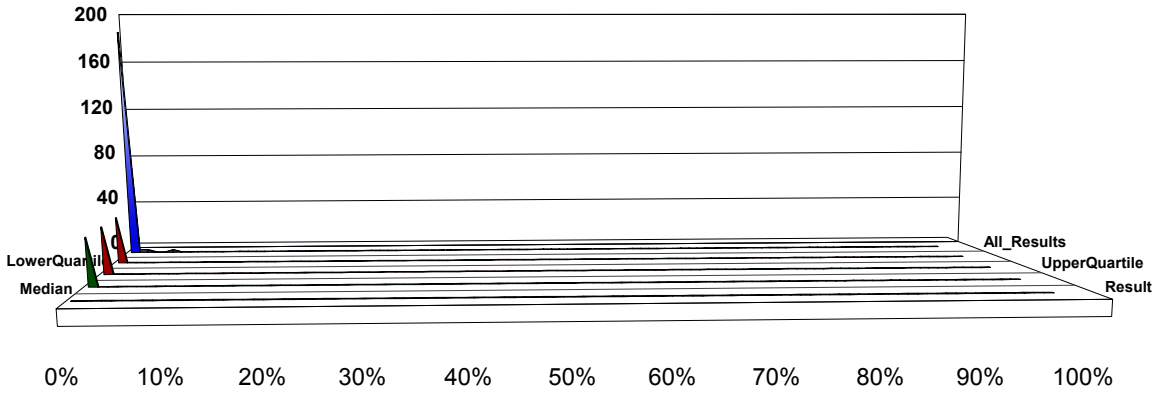
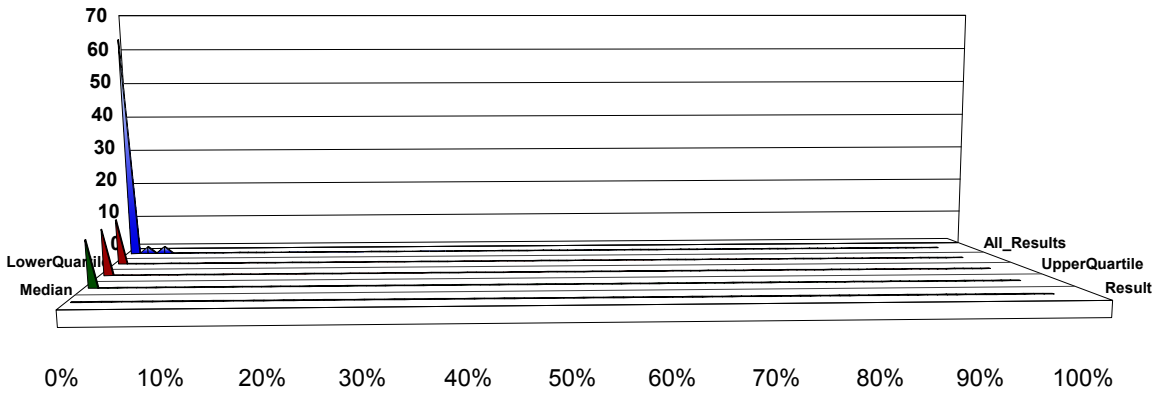
Granulocytes PNH Clone - FLAER



Granulocytes PNH Clone - CD24



Granulocytes PNH Clone - CD16



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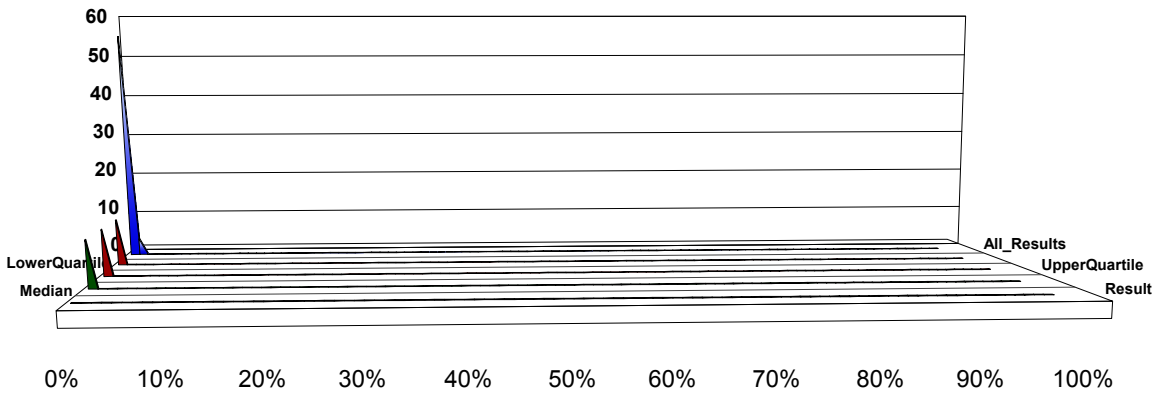
Granulocytes PNH Clone - CD66b



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Method Breakdown

RBC PNH Clone

Flow Cytometer	Median	Lower Quartile	Upper Quartile	Returns
CD55				
All	0.00	0.00	0.10	22
BD	0.00	0.00	0.10	18
CD59				
All	0.00	0.00	0.00	122
BD	0.00	0.00	0.00	84
Coulter	0.00	0.00	0.00	37

Antibody Manufacturer	Median	Lower Quartile	Upper Quartile	Returns
CD55				
All	0.00	0.00	0.10	22
BD Biosciences	0.05	0.00	0.10	12
CD59				
All	0.00	0.00	0.00	122
BD Biosciences	0.00	0.00	0.34	12
Beckman Coulter	0.00	0.00	0.03	12
eBioscience	0.00	0.00	0.00	10
invitrogen	0.00	0.00	0.00	60

Fluorochrome	Median	Lower Quartile	Upper Quartile	Returns
CD55				
All	0.00	0.00	0.10	22
PE	0.05	0.00	0.53	16
CD59				
All	0.00	0.00	0.00	122
PE	0.00	0.00	0.00	95
FITC	0.00	0.00	0.05	27

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Method Breakdown
Granulocytes PNH Clone

Flow Cytometer	Median	Lower Quartile	Upper Quartile	Returns
CD157				
All	0.00	0.00	0.00	10
CD16				
All	0.00	0.00	0.00	35
BD	0.00	0.00	0.00	25
CD24				
All	0.00	0.00	0.00	97
BD	0.00	0.00	0.00	65
Coulter	0.00	0.00	0.00	31
CD55				
All	0.00	0.00	0.07	19
BD	0.00	0.00	0.10	11
CD59				
All	0.00	0.00	0.00	20
BD	0.00	0.00	0.03	12
CD66b				
All	0.00	0.00	0.00	30
BD	0.00	0.00	0.00	22
FLAER				
All	0.00	0.00	0.00	122
BD	0.00	0.00	0.00	82
Coulter	0.00	0.00	0.00	39

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Antibody Manufacturer	Median	Lower Quartile	Upper Quartile	Returns
CD157				
All	0.00	0.00	0.00	10
CD16				
All	0.00	0.00	0.00	35
BD Biosciences	0.00	0.00	0.00	13
Beckman Coulter	0.00	0.00	0.00	13
CD24				
All	0.00	0.00	0.00	97
BD Biosciences	0.00	0.00	0.00	23
Beckman Coulter	0.00	0.00	0.00	45
Pharmingen	0.00	0.00	0.00	20
CD55				
All	0.00	0.00	0.07	19
CD59				
All	0.00	0.00	0.00	20
CD66b				
All	0.00	0.00	0.00	30
BD Biosciences	0.00	0.00	0.00	13
FLAER				
All	0.00	0.00	0.00	122
Other	0.00	0.00	0.00	66
Protox	0.00	0.00	0.00	46

Fluorochrome	Median	Lower Quartile	Upper Quartile	Returns
CD157				
All	0.00	0.00	0.00	10
CD16				
All	0.00	0.00	0.00	35
CD24				
All	0.00	0.00	0.00	97
PE	0.00	0.00	0.00	71
CD55				
All	0.00	0.00	0.07	19
PE	0.00	0.00	0.03	13
CD59				
All	0.00	0.00	0.00	20
PE	0.00	0.00	0.00	13
CD66b				
All	0.00	0.00	0.00	30
FITC	0.00	0.00	0.00	22
FLAER				
All	0.00	0.00	0.00	122
FITC	0.00	0.00	0.00	34
AlexaFluor 488	0.00	0.00	0.00	87

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PNH 151601

Sample 077

This sample was PNH material spiked into a unit of normal whole blood; the median values of the results returned accurately reflected those obtained at pre-issue testing.

The consensus result for this sample was **Clone Present** for both Red Cell and Granulocyte clones. 2 centres did not identify a Red Cell PNH clone upon analysis, with one centre recording a 0% clone as clone absent and the other recording it as clone present. 2 centres identified clones of 0.3% and 1.5% recording these as clone absent. 2 other centres did not submit any information in relation to PNH clone size but specified clone present. Participants are reminded to submit both PNH clone size and to specify Clone Present or Clone Absent.

Only 2 participants failed to identify the granulocyte clone on all of their markers, and reported their results as being clone absent. Again, 2 other centres did not submit any information in relation to PNH clone size but specified clone present.

7 centres did not return any results for this sample.

Sample 078

This sample was a normal whole blood sample and contained no PNH clone.

The consensus result for this sample was **Clone Absent** for both Red Cell and Granulocyte clones. 24 participants identified a red cell clone and 20 of these were reported as clone absent (clone size 0.01%-0.60%), the remaining 4 centres reported the results (0.05%, 2.14%, 4.20% and 18.0%) as being clone present. 2 other centres did not submit any information in relation to PNH clone size but specified clone absent.

21 participants recorded a granulocyte clone on at least one of their markers, 17 of these were reported as being clone absent (clone size 0.01% to 36.84%), whilst 4 centres reported the results as clone present (0.02%-5.8%). 3 other centres did not submit any information in relation to PNH clone size but specified clone absent.

7 centres did not return results for this sample.

Please note that UK NEQAS for Leucocyte Immunophenotyping will be running Instrument specific PNH testing courses throughout the year, details of which are included at the end of this report.

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Information with respect to compliance with standards BS EN ISO/IEC 17043:2010

4.8.2 a) The proficiency testing provider for this programme is:
UK NEQAS for Leucocyte Immunophenotyping
Pegasus House, 4th Floor Suite
463A Glossop Road
Sheffield, S10 2QD
United Kingdom
Tel: +44 (0) 114 267 3600, Fax: +44 (0) 114 267 3601
e-mail: nicola.rose@ukneqasli.co.uk

4.8.2 b) The coordinators of UK NEQAS LI programmes are Prof David Barnett and Mr Liam Whitby.

4.8.2 c) Person(s) authorizing this report:
Prof David Barnett, Director or Mr Liam Whitby, Operations Manager of UK NEQAS LI

4.8.2 d) No activities in relation to this EQA exercise were subcontracted.

4.8.2 g) The UK NEQAS LI Confidentiality Policy can be found in the Quality Manual which is available by contacting the UK NEQAS LI office. Participant details, their results and their performance data remain confidential unless revealed to the relevant NQAAP when a UK participant is identified as having performance issues.

4.8.2 i) All EQA samples are prepared in accordance with strict Standard Operational Procedures by trained personnel proven to ensure homogeneity and stability. Where appropriate/possible EQA samples are tested prior to issue. Where the sample(s) issued is stabilised blood or platelets, pre and post stability testing will have proved sample suitability prior to issue.

4.8.2 l), n), o), r) & s) Please refer to the UK NEQAS LI website at www.ukneqasli.co.uk for detailed information on each programme including the scoring systems applied to assess performance. Where a scoring system refers to the 'consensus result' this means the result reported by the majority of participants for that trial issue. Advice on the interpretation of statistical analyses and the criteria on which performance is measured is also given. Please note that where different methods/procedures are used by different groups of participants these may be displayed within your report, but the same scoring system is applied to all participants irrespective of method/procedure used.

4.8.2 m) We do not assign values against reference materials or calibrants.

4.8.2 q) Details of the programme designs as authorized by The Steering Committee and Specialist Advisory Group can be found on our website at www.ukneqasli.co.uk. The proposed trial issue schedule for each programme is also available.

4.8.2 t) If you would like to discuss the outcomes of this trial issue, please contact UK NEQAS LI using the contact details provided.

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Directions

Sheffield Hallam University is based at the heart of the country in Britain's fifth largest city. Major road and rail networks link directly with the rest of the country and the city itself is served by excellent bus and Supertram services. The University's city campus is directly adjacent to Sheffield Central Passenger Transport Interchange at Pond Street, and is a two minute walk from Sheffield Midland railway station.

Rail Links
Regular intercity services operate between Sheffield and other major towns and cities. The journey from London takes around two hours. Manchester, Leeds and Birmingham are an hour away.

Road Links
Sheffield is easily accessible by road with a dual carriageway link (A630) from junction 33 of the M1 directly into the city centre.

The Route to the University:

- From junction 33 of the M1, follow signs for A630 Sheffield City Centre.
- Continue on the A630 until the dual carriageway ends at a large roundabout with traffic signals (Park Square).
- Take the fourth exit from the roundabout signposted A61 Chesterfield.
- Follow this road until the next roundabout (Furnival Square).
- Take the first left, then the second left onto Arundel Street.
- The entrance to the university is straight ahead on Howard Street.

There are NCP car parks on Arundel Gate, Eyre Street and Arundel Street. There is also limited on-street pay and display parking within proximity of the venue.

How to register

For full details on how to register please visit our website **www.ukneqasli.co.uk**

UK NEQAS for Leucocyte Immunophenotyping

Tel: +44 (0)114 267 3600
Fax: +44 (0)114 267 3601
Email: nicola.rose@ukneqasli.co.uk
www.ukneqasli.co.uk

You may also be interested in…

NACFC
NATIONAL ACADEMY FOR CLINICAL FLOW CYTOMETRY

Dedicated courses for PNH detection by flow cytometry.

See **www.sheffield.ac.uk/nacfc** or call **0114 267 3600** for further details.

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UK NEQAS
Leucocyte Immunophenotyping

Scientific Meeting

Monday, 29th June 2015

Pennine Suite
Sheffield Hallam University
City Campus
Sheffield
S1 1WB

2015 Programme

2015 Programme

UK NEQAS for Leucocyte Immunophenotyping Scientific Meeting.

8.30am - 9.15am **Registration and Coffee**

9.15am - 9.20am **Welcome Address** PENNINE THEATRE

Professor David Barnett, Director, UK NEQAS for Leucocyte Immunophenotyping, Sheffield

9.20am - 10.10am **Plenary Lecture 1** PENNINE THEATRE

The Impact of The Barnes Report

Speaker: Dr David Hughes, Royal Hallamshire Hospital, Sheffield, UK

Chair: Mr Neil Porter, Chairman of the UK NEQAS for Leucocyte Immunophenotyping Steering Committee, Sheffield, UK

10.10am - 11.00am **Plenary Lecture 2** PENNINE THEATRE

Flow Cytometry and Molecular Diagnostic in the Clinical Laboratory: What Really Is Best Practice?

Speaker: Professor David Barnett

Chair: Mr Neil Porter

11.00am - 11.30am **Coffee Break & Trade Exhibitions**

11.30am - 12.15pm **Plenary Lecture 3** PENNINE THEATRE

What Do I Need to Pass ISO 15189: An End User Experience?

Speaker: Mr David Bloxham, Addenbrookes Hospital, Cambridge, UK

Chair: Professor Thomas Denny, Chief Operating Officer, Duke Human Vaccine Institute and Center for HIV/AIDS Vaccine Immunology and Director, Immunology and Virology Quality Assessment Center, Durham, USA

12.15pm - 1.15pm **Lunch & Trade Exhibitions**

1.15pm - 2.45pm **Short Presentations**

Presentation A: Immunophenotyping PENNINE THEATRE

Establishing Competency of Staff in Flow Cytometry

Speaker: Mr Andrew Moran, Central Manchester University Hospitals NHS Foundation Trust, Manchester, UK

How to Establish a Multi Colour Flow Cytometry Panel

Speaker: Dr Ruth de Tute, St James Hospital, Leeds, UK

How to Diagnose AML and MDS by Flow Cytometry: ICSH Guidelines for Flow Cytometric Evaluation of Patients with Suspected Acute Myeloid Leukaemia and Myelodysplastic Syndromes

Speaker: Professor Wolfgang Kern Munich, Germany

Chair: Professor David Barnett

Presentation B: Molecular PEAK THEATRE

Next Generation Sequencing in Haematological Malignancy: A European Perspective

Speaker: Professor Wolfgang Kern, Munich, Germany

Next Generation Sequencing in Haematological Malignancy: A UK Perspective

Speaker: Dr Nicholas Lea, Kings College Hospital, London

Applications of NGS in the diagnosis of lymphoid malignancies

Speaker: Mrs Dörte Wren, The Royal Marsden NHS Foundation Trust, London, UK

Chair: Professor John Snowden, Consultant Haematologist & Director of BMT, Royal Hallamshire Hospital, Sheffield, UK

2.45pm - 3.05pm **Coffee Break**

3.05pm - 4.05pm **UK NEQAS LI Focus Sessions**

Session A: Immunophenotyping PENNINE THEATRE

Chair: Mr Neil Porter

To be conducted by UK NEQAS LI staff and SC members on the following topics:

- *CAPA Process/RCA*
- *Nomenclature*
- *Sample Handling*

Session B: Molecular PEAK THEATRE

Chair: Dr Chris Dalley, Consultant Haematologist, The Royal Sussex County Hospital, Brighton, UK

To be conducted by UK NEQAS LI staff and SC members on the following topics:

- *CAPA Process/RCA*
- *Nomenclature*
- *Sample Handling*

4.05pm **Coffee and Depart**

This meeting is registered for IBMS and RCPath CPD points

NACFC

NATIONAL ACADEMY
FOR CLINICAL FLOW CYTOMETRY

UK NEQAS for Leucocyte Immunophenotyping (UK NEQAS LI) based in the Royal Hallamshire Hospital, Sheffield Teaching Hospitals NHS Foundation Trust, the University of Sheffield and Alexion (UK) are proud to announce the formation of the National Academy of Clinical Flow Cytometry.

This facility has been founded on an unrestricted educational grant from Alexion (UK) and will be based at the University of Sheffield. The aims of the Academy are to provide tuition from international experts to Healthcare Scientists performing clinical flow cytometric analysis and to ensure that these tests are performed at the highest standard. The academy will therefore have a positive benefit for the treatment and management of thousands of patients across the UK and beyond.

Paroxysmal Nocturnal Haemoglobinuria (PNH) testing course

This specific course aims to train scientists in the flow cytometric detection and monitoring of Paroxysmal Nocturnal Haemoglobinuria (PNH). It is a rare disease, occurring at any age, with a prevalence of 2 cases per million and can be fatal if not detected and treated correctly.

This initiative will aim to improve detection and monitoring of PNH in clinical flow cytometry laboratories within the National Health Service.

It is envisaged that by improving both knowledge and skills of healthcare scientists there will be a dramatic improvement in the diagnosis and monitoring of PNH. This will lead to improved standardization of testing procedures at a National and ultimately International level.

Faculty

The National Academy, led by Professor David Barnett and his UK NEQAS LI team, will be based within the University of Sheffield. It has a faculty of internationally respected scientists such as Professor Robert Sutherland (Toronto, Canada), Andrea Illingworth (Bangor, USA) and Dr Steven Richards (HMDS, Leeds) who will teach clinical healthcare scientists practicing in UK hospitals on how to implement best practices for PNH testing.

Course details

Each course will be specifically targeted at an instrument type currently in use for clinical flow cytometry and will be delivered over 3 days by international experts in the field. All courses will feature classroom and practical sessions that will ensure that students receive the best tuition in the flow cytometric detection of PNH.

Registration

Via on-line form on our website at **www.sheffield.ac.uk/nacfc**

Registration for the course costs £100 and includes 2 nights' accommodation, all meals and pre- and post-course practical assessments.

Course dates

Becton Dickinson
Flow Cytometer Course
13–15 July
(BD FACSCalibur and BD FACSCanto II Practical Sessions)
Tutors: Prof. Rob Sutherland, Dr Steven Richards, Daniel Payne

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Beckman Coulter
Flow Cytometer Course
21–23 September
(BC FC500 and BC Navios Practical Sessions)
Tutors: Andrea Illingworth

Becton Dickinson
Flow Cytometer Course
26–28 October
(BD FACSCalibur and BD FACSCanto II Practical Sessions)
Tutors: Prof. Rob Sutherland, Daniel Payne

Beckman Coulter
Flow Cytometer Course
23–25 November
(BC FC500 and BC Navios Practical Sessions)
Tutors: Daniel Payne, Andrea Illingworth

NACFC

Sponsor

ALEXION

Jointly hosted by

UK NEQAS
Leucocyte Immunophenotyping

The University Of Sheffield.

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